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Using Orchardgrass and Endophyte-Free Fescue Versus Endophyte-Infected Fescue Overseeded on Bermudagrass for Cow Herds: I. Four-Year Summary of Forage Characteristics

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ABSTRACT

A systems trial was designed to evaluate forage characteristics within mixed-species pastures consisting of (i) endophyte-infected tall fescue (*Festuca arundinacea* Schreb.; E+) mixed with common bermudagrass [*Cynodon dactylon* (L.) Pers.] and other forages; (ii) endophyte-free tall fescue (E−) overseeded into dormant common bermudagrass; and (iii) orchardgrass (OG; *Dactylis glomerata* L.) established under the same conditions as E−. The E− and OG pastures were grazed with either twice weekly (2W) or twice monthly (2M) rotation schedules, but E+ was grazed only as 2M. Across 41 sampling dates (2000 through 2003) the mean forage mass across all forage systems was 3809 kg ha^{-1}, and there was an interaction of forage system and sampling date ($P = 0.001$). In vitro dry matter disappearance (IVDMD) and crude protein (CP) varied ($P < 0.0001$) with sampling date in seasonal patterns that were generally predictable. Frequencies of tall fescue in E− and E+ pastures increased ($P < 0.10$) over years, reaching numerical maxima (61 to 72%) at the end of the trial. For OG, frequencies reached numerical maxima of 52 and 42% in 2W and 2M pastures, respectively, but then declined ($P < 0.10$) over time, ending at 39 and 24%, respectively. At the end of the trial, reinfection of OG pastures by rogue E+ plants reached a numerical maximum frequency of only 10%, and concentrations of total ergot alkaloids in tall fescue plants from E− pastures were only about 25% of those for E+ pastures, thereby suggesting that pasture toxicity can be reduced substantially for at least 5 yr using these alternative forage systems.

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The association of the fungus *Neotyphodium coenophialum* (Morgan-Jones and Gams) Glenn, Bacon, and Hamlin comb. nov. (Glenn et al., 1996) with tall fescue has a positive effect on plant growth and persistence (Siegel et al., 1985), but the toxins produced by this fungus affect livestock performance negatively (Read and Camp, 1986; Peters et al., 1992; Schmidt and Osborn, 1993; Paterson et al., 1995). These problems cost livestock producers in the USA an estimated $609 million annually (Hoveland, 1993). Although livestock performance is affected negatively, the symbiotic association of the endophyte with tall fescue plants is particularly important with respect to survival and persistence of tall fescue throughout the southern Ozarks. Endophyte-infected plants have physiological mechanisms that permit better growth, survival, and competitiveness under stressful growing conditions (Hill et al., 1991; Bouton et al., 1993; West et al., 1993; Malinowski and Belesky, 2000) than plants not infected with an endophyte. Many Ozark pasture soils are shallow, have poor water-holding capacity, and are often acidic with relatively low fertility (Sauer et al., 1998). In addition, growing conditions are compounded further by (i) relatively high mean daily temperatures during summer (27.9 and 27.2°C, during July and August, respectively, at Batesville, AR; NOAA, 2002); (ii) species competition from bermudagrass and other perennial warm-season grasses; and (iii) a frequent habit of overgrazing by many cow-calf producers throughout this region. These factors combine to create an extremely stressful environment for tall fescue, and many mixed species pastures on producer farms contain 50% tall fescue or less. Other perennial cool-season grasses that possess no endophyte, such as E− and OG, may support better animal performance, but generally have persisted poorly in the southern Ozarks when subjected to the same harsh conditions and management as E+.

Dilution of E+ pastures with other nontoxic grasses or legumes has improved both cow (Holloway and Butts, 1984; Gay et al., 1988; Waller et al., 1989) and stocker cattle performance (Coffey et al., 1990; McMurphy et al., 1990; Chestnut et al., 1991), but dilution of E+ pastures has rarely offset all of the performance reductions associated with tall fescue toxicosis. While competition with common bermudagrass creates an additional stressor for tall fescue, common bermudagrass and various other grasses and legumes also can provide

Abbreviations: CP, crude protein; DM, dry matter; E+, endophyte-infected tall fescue; E−, endophyte-free tall fescue; IVDMD, in vitro dry matter disappearance; OG, orchardgrass; 2M, rotation to new paddocks twice monthly; 2W, rotation to new paddocks twice weekly.

a measure of natural dilution within many southern Ozark pastures. Throughout the southern Ozarks, the relative proportions of E+ and bermudagrass in mixed-species pastures that optimize cattle performance are unknown, and likely vary with precipitation, temperature, fertilization, and many other factors. To some extent, relative proportions of tall fescue and common bermudagrass can be managed to the advantage of one species or the other by the amount and timing of N fertilization, as well as by controlling closely the timing, frequency, and height of mowing or grazing (Wilkinson et al., 1968; Hoveland et al., 1978; Fribourg and Overton, 1979; Pitman, 1999). While it is unclear what relative proportions of E+ and common bermudagrass may optimize cattle performance, both species are likely important; E+ extends the grazing season by providing forage in the spring and fall when temperatures are cool, while bermudagrass grows actively throughout the summer and dilutes the concentrations of toxins produced within E+ plants.

Another option available to cow-calf producers in the southern Ozarks is to convert their acreage to fescue varieties containing novel, non-ergot-alkaloid-producing endophytes (Bouton et al., 2002; Nihsen et al., 2004). While promising, producers are not likely to convert all, or even a majority of their pastures to these novel endophyte–fescue associations because of expense and/or topography. However, other management options may be effective in reducing the effects of fescue toxicosis for cow-calf producers. One option would be to apply glyphosate [*N*-(phosphonomethyl)glycine] to mixed species pastures, thereby killing existing cool-season plants and releasing a base of common bermudagrass. Nontoxic forages, such as E− and OG could then be established by sod-seeding techniques, which producers in the region understand and have suitable equipment to carry out. This approach would not completely kill all existing (and presumably E+) tall fescue plants (Defelice and Henning, 1990), and reinfection of the pasture by surviving rogue E+ plants would likely occur. However, a spring or fall application of glyphosate would release the base of common bermudagrass within these pastures, thereby retaining considerable pasture productivity until OG or E− could be established. Furthermore, E− and OG have much lower seed costs than novel-endophyte tall fescues.

This management approach has several obvious uncertainties. Because of the potential for reinfection of pastures with E+ plants, this management option cannot be considered permanent. It is unclear how much livestock performance may be improved or how long this potentially higher level of performance may continue before it is limited by reinfection of pastures by E+ plants, poor persistence of E− or OG forages, increased frequency of bermudagrass, or various combinations of these potential occurrences. Persistence of E− overseeded into bermudagrass was improved in central Georgia with a 12-paddock rotation compared to continuous stocking (Hoveland et al., 1997). However, it is uncertain if rotational management will improve persistence of E− in the southern Ozark Highlands. Also, to the best of our knowledge, factors affecting the competitive balance between OG and common bermudagrass have not been evaluated, probably because OG is less tolerant of heat (Baker and Jung, 1968) and drought than tall fescue, and stands in the South generally do not persist more than 2 to 4 yr (Ball et al., 2002). Therefore, a 4-yr trial was initiated in January 2000 to evaluate the effectiveness of E− or OG overseeded into dormant common bermudagrass sods for spring-calving cows. Our objective was to evaluate forage characteristics for these nontoxic (E− or OG) forages at two rotational frequencies (2M or 2W), and to compare these forage systems with a typical pasture mixture of approximately 50% E+, plus common bermudagrass and other forages that is representative of those observed commonly throughout the southern Ozarks.

MATERIALS AND METHODS

Establishment and Management of Experimental Pastures

Location

This study was conducted at the Batesville Livestock and Forestry Branch Station (35°50′ N, 91°48′ W), located near Batesville, AR. The experimental site covered 52 total ha, and was divided into thirteen 4-ha pastures. The soil types found within the pasture area consisted primarily of a Gepp very cherty silt loam (very-fine, mixed, mesic Typic Paleudalf) with 3 to 12% slopes, or a Noark very cherty silt loam (clayey-skeletal, mixed, mesic Typic Paleudult) with 8 to 12% slopes.

Establishment of Pastures

Before initiating this study, the experimental pastures had been used for a variety of grazing and/or supplementation studies; pastures generally contained 50% E+ or less and had a vigorous base sod of common bermudagrass, which is not unusual for mixed species pastures throughout the region. In the spring of 1997, nine of the 13 existing 4-ha pastures were sprayed with glyphosate at a rate of 1.68 kg a.i. ha^{-1} to eliminate annual and perennial cool-season grasses. The species composition of the remaining four pastures was mixed, but each contained approximately 50% E+ and a base sod of common bermudagrass; these pastures were retained as controls. Glyphosate was applied to each of the nine pastures only once because we felt this represented the likely limit of investment that many producers would be willing to make for eradication of E+ on marginally productive land. In addition, it was not a research goal to eliminate bermudagrass, which would have been damaged by multiple applications during the growing season. These nine pastures were then grazed as bermudagrass pastures during the summers of 1997 and 1998, and were used in another grazing study that required overseeding with cereal rye (*Secale cereale* L.) and/or annual ryegrass (*Lolium multiflorum* Lam.) in the fall of 1997.

All experimental pastures were grazed intermittently throughout the early summer of 1998. Beginning in August 1998, mob-grazing management was used to remove the existing summer forage growth from the nine pastures previously sprayed with glyphosate; at that time, the existing forage was primarily common bermudagrass. Mob-grazing techniques were used because several of the pastures contained rock outcroppings or other areas where the terrain was rough; therefore, they were unsuitable for haying. In late September and early October 1998, 'Kentucky 31' E− and 'Benchmark' OG were overseeded into their assigned pastures at respective rates of 26.5 and 24.7 kg pure live seed ha^{-1}. All seeding was completed by 9 Oct. 1998. Before establishment, a randomly collected sample of E− seed was submitted for determination of endophyte-infection percentage (Fescue Diagnostic Center, Auburn University, Auburn, AL), and found to be 1% endophyte infected. Pastures were established with a 2.1-m-wide Tye Pasture Pleaser drill (The Tye Company, Lockney, TX) with rows spaced at 25 cm. Soil tests taken before establishment indicated that fertility levels for P, K, and lime varied only minimally from the soil test recommendations of the Arkansas Cooperative Extension Service (Chapman, 2001); therefore, any needed applications of P and K were deferred until 24 Feb. 1999. All pastures were fertilized with urea (46–0–0) at a rate of 50 kg N ha^{-1} on 18 Feb. 1999.

In April 1999, three independent observers evaluated each pasture overseeded with OG or E− visually for continuous row coverage by cool-season seedlings. Observers walked each pasture in a zig-zag pattern, covering the entire pasture area before making their estimate. These estimates were averaged for each pasture before statistical analysis. During the 1999 growing season, overseeded pastures were grazed lightly to control forage growth and to allow new seedlings a chance to become well established.

Maintenance of Pastures

On 9–10 Sept. 1999, all pastures were fertilized with urea (46–0–0) at a rate of 67 kg N ha^{-1}; similar applications were made each subsequent year in mid February, early June, and early September. Therefore, a total of 202 kg N ha^{-1} were applied annually after cattle were allocated and grazing was initiated. Soil tests were obtained each year in August and any needed P, K, or lime was applied each September based on soil test recommendations (Chapman, 2001). Broadleaf weeds were controlled as necessary throughout the trial with picloram (4-amino-3,5,6-trichloropicolinic acid, triisopropanolamine salt) and 2,4-D (2,4-dichlorophenoxyacetic acid, triisopropanolamine salt) applied in combination at respective rates of 0.3 and 1.1 kg a.i. ha^{-1}.

Pasture Rotation Schedules

Experimental pastures were grazed with two rotational grazing schemes (Table 1) that included rotations to fresh paddocks either 2W or 2M. Pastures grazed with the 2W rotation frequency were subdivided into eight 0.5-ha paddocks that were grazed for 3 to 4 d during each rotation, and then rested for the balance of the month (26–28 d). Pastures grazed with the 2M rotation frequency were subdivided into two 2.0-ha paddocks. Cows assigned to these pastures were maintained on a specific paddock for 15 d before they were rotated to the other paddock for the remainder of the month. The OG and E− pastures were grazed with both 2W and 2M schedules, largely because it was hypothesized that rotation frequency may strongly affect the persistence of nontoxic E− and OG plants. The E+ control pastures were typical of mixed pastures of E+ and common bermudagrass throughout the region; these were grazed with the 2M rotation frequency only because that represented the likely upper limit of time and management that most part-time cow-calf producers would be willing to invest for pastures with E+.

Cattle Management

All specific details describing cattle management are discussed in depth in a companion report (Coblentz et al., 2006). Briefly, 65 spring-calving cows (548 ± 68.0 kg) were stratified by weight, age, and breeding and assigned to each of the 13 pastures (five per pasture) on 11 Jan. 2000. From mid-May through mid-July of each year, one Gelbvieh bull was assigned to each experimental pasture. Cows remained on their assigned pasture, without additional randomization between years, for the entire 4-yr trial, and were replaced only if they died, were confirmed open, or did not produce a live calf. Cows and calves were not supplemented with concentrate feeds at any time; however, a commercial mineral mix was offered free choice.

Whenever available forage was limiting, cows and calves were confined within a single 2W paddock (0.5 ha) or an area of comparable size constructed with temporary electric fencing within a 2M paddock and offered bermudagrass hay harvested from another location on the research station in round-bale feeders on an ad libitum basis. These 0.5-ha feeding areas were not relocated during the trial. These feeding techniques were used to prevent continued grazing over the entire pasture area when forage was limiting, and to confine both tractor and cattle traffic to a small, but convenient, area. The decision to feed hay was based on either of two criteria: (i) when forage mass dropped below 1000 kg ha^{-1} as measured by a calibrated disk meter (Bransby et al., 1977); or (ii) when the grazed stubble height of the appropriate cool-season grass (OG, E−, or E+) dropped below 7.5 cm. The second decision trigger was necessary, especially in the late fall, because cows grazed cool-season forages (particularly OG) preferentially over frosted bermudagrass.

In an effort to control the flush of forage growth that occurs in the spring, extra "thin" fall-calving cows were assigned to each pasture to improve their body condition. This technique was used because all pastures were not suitable for measuring

Table 1. Description of grazing systems evaluated near Batesville, AR, from 2000 through 2003. Mixed species pastures contained common bermudagrass with either endophyte-free tall fescue (E−), orchardgrass (OG), or endophyte-infected tall fescue (E+). The OG and E− pastures were grazed with either a twice weekly (2W) or twice monthly (2M) rotation schedule, while the E+ pastures were grazed with a 2M schedule only.

Cool-season perennial forage	Rotation schedule	Replications	Total pasture size	No. paddocks per pasture	Paddock size	Rotation frequency	Rest period†	Base stocking rate‡
		no.	ha	no.	ha	d	d	pairs ha^{-1}
OG	2W	3	4.0	8	0.5	3–4	26–28	1.25
	2M	2	4.0	2	2.0	15	15	1.25
E−	2W	2	4.0	8	0.5	3–4	26–28	1.25
	2M	2	4.0	2	2.0	15	15	1.25
E+	2M	4	4.0	2	2.0	15	15	1.25

† Rest period for each paddock between grazing events.

‡ Pastures were stocked permanently with five spring-calving cows. Calves were weaned from these cows in early October of each year. To control the flush of spring forage growth each year, thin fall-calving cows were added to each pasture system. Tabulation of grazing days, weight gains, and body condition scores for these extra cows are reported separately (Coblentz et al., 2006). The reported stocking rate reflects only the permanently assigned spring-calving cows and their calves.

any extra forage produced as hay. All details of these procedures and summaries of performance for these cows are described in detail within in a companion report (Coblentz et al., 2006).

Analytical Procedures and Measurements

Pasture Measurements

Except during the winter months when cows were offered hay, pastures were evaluated monthly for forage mass using a calibrated disk meter (Bransby et al., 1977). This was accomplished by walking each pasture in a zig-zag pattern and then estimating forage mass at 6 locations ha^{-1} (24 locations per pasture). Simultaneously, forage samples were collected at half (12 per pasture) of these locations by clipping forage to a 2.5-cm stubble height with hand shears. All forage samples were dried to a constant weight under forced air at 50°C, and then ground through a Wiley mill (Arthur H. Thomas, Philadelphia, PA) fitted with a 1-mm screen before determination of IVDMD and CP. On each sampling date, all disk meter readings and associated forage samples clipped for analysis of nutritive value were obtained from sites distributed uniformly over the entire 4-ha pasture.

Pastures were evaluated for basal cover and species composition before initiating the trial (November 1999) by the modified step-point method (Owensby, 1973). Each pasture was walked in a zig-zag pattern placing the step-pointer on the ground at 40 locations ha^{-1} (160 per pasture). At each placement of the step-pointer, two observations were recorded: (i) whether the pointer directly touched the base of a plant; and (ii) the plant species either touching the pointer directly, or the nearest species within an 180° arc in front of the pointer. Total basal cover was calculated as the percentage of pointer placements that directly touched the base of any plant. Species frequency was calculated as the percentage of total placements that a particular species was either touched directly or was closest to the pointer. These procedures were repeated in June and November of each subsequent year to assess the cumulative effects of grazing on the species composition and basal cover within each pasture. The June evaluation date corresponded generally to the time period of peak forage mass for cool-season grasses. Conversely, the November evaluation date was scheduled immediately after the onset of fall dormancy to reflect bermudagrass that had accumulated throughout the summer months. By this time, below-freezing temperatures had caused the bermudagrass to turn brown, which made it easy to distinguish from cool-season grasses that were still growing actively. To simplify the presentation of results and subsequent discussion, June and November evaluations within each calendar year were averaged, thereby creating an annual mean for each pasture that was used for all statistical analyses of species composition data.

In 2000 and 2001, it was relatively easy to distinguish rogue (and presumably E+) tall fescue plants within E− pastures that were not killed by the application of glyphosate in 1997. This differentiation was based on large differences in crown size and spatial orientation of rogue plants relative to the drill rows. After the 2001 grazing season, these distinctions could not be made with any confidence, and separate identification of rogue fescue plants was discontinued. During all evaluations of species frequency, tall fescue plants were identified only visually; no attempt was made to establish the endophyte status of each plant by independent laboratory testing.

In June of each year, additional samples of tall fescue forage were obtained to quantify concentrations of total ergot alkaloids. These samples were collected by walking each pasture in a zig-zag pattern, and clipping tall fescue plants to a 2.5-cm stubble height with hand shears from approximately 4 random locations ha^{-1} (16 locations per pasture). Samples were limited to tall fescue forage only; all other species were removed from the sample. After collection, tall fescue samples were sealed immediately in plastic freezer bags, and then submerged in ice in an insulated cooler. Each hour during collection, samples were transported to a conventional freezer (−4°C) and stored for not more than 48 h before they were transported to Fayetteville on ice and then stored in an ultra-low temperature freezer (−80°C). Samples were then lyophilized, ground through a 1-mm screen as described previously, and then returned to the ultra-low freezer until they were analyzed for total ergot alkaloids. Only rogue fescue plants were sampled from E− pastures in 2000 and 2001. After that time, rogue plants could not be identified with confidence, and all fescue plants were sampled at random. Within some E− pastures during 2000 and 2001, species frequency estimates for rogue fescue plants were very low or 0%. If technicians sampling these pastures for total ergot alkaloids could not find rogue plants to sample, then all fescue plants were sampled at random to provide independent, corroborative evidence of a low rate of endophyte infection and concomitant low toxicity within these pastures.

Laboratory Analyses

Dried and ground forage samples were analyzed for total N by rapid combustion (AOAC, 1998; AOAC Official Method 990.93; Elementar Americas, Inc., Mt. Laurel, NJ), and for IVDMD by the batch techniques outlined by ANKOM Technology Corp. (Fairport, NY). Concentrations of CP within each forage were calculated by multiplying the percentage of total N in each forage sample by 6.25. Rumen fluid for IVOMD analysis was obtained from two ruminally cannulated crossbred steers that were offered a diet consisting of 850 g kg^{-1} alfalfa hay and 150 g kg^{-1} concentrate at a daily rate of 20 g kg^{-1} of BW (as-is basis). On an as-is basis, the concentrate mix contained 910 g kg^{-1} cracked corn, 40 g kg^{-1} liquid molasses, and 50 g kg^{-1} trace mineral salt. The steers were adapted to the diet for a minimum of 7 d before collecting the rumen fluid. Total ergot alkaloids in tall fescue forages were quantified using the procedures of Hill and Agee (1994; Agrinostics, LLC, Athens, GA).

Statistical Analyses

Individual pastures served as the experimental unit for all statistical analyses. Forage mass and concentrations of IVDMD and CP were analyzed as a split-plot design with forage system as the whole-plot term, and the 41 sampling dates as the subplot (repeated measures) term. The whole-plot term (forage systems) was arranged in a completely randomized design and tested for significance using the pasture within forage system mean square as the error term. Sampling date, and the forage system × sampling date interaction were tested for significance with the residual error mean square.

Species composition, basal cover, and concentrations of total ergot alkaloids were all analyzed as a split-plot (repeated measures) design with forage systems as the whole-plot term and year as the subplot or repeated measures term. In each of these cases, forage system was tested for significance as described previously; the repeated measures and interaction terms were tested with the residual error mean square. All statistical analyses were conducted with PROC GLM of SAS (SAS Institute, 1989). Generally, least square means were separated with the PDIFF option; however, main-effect means of sampling date were balanced, thereby permitting calculation of LSD. Statistical significance was declared at $P < 0.10$, unless otherwise noted.

RESULTS AND DISCUSSION

Precipitation

Generally, annual rainfall during the 4-yr trial (Table 2) can best be described as one relatively dry year (2000), two nearly normal years (2001 and 2002), and one relatively wet year (2003). During July and August 2000, there was only a cumulative total of 27 mm of rainfall, which created extreme drought stress and necessitated supplementation of cows and calves with hay.

Establishment

The visual assessment of continuous drill row coverage by seedling E− and OG plants in April 1999 indicated there were no initial differences ($P \geq 0.615$) in establishment across OG2W, OG2M, E-2W, and E-2M pastures. The overall mean of 68% (data not shown) suggests that initial establishment was relatively good considering the rough and rolling terrain, shallow soils, and competition from vigorous stands of common bermudagrass that comprised the vast majority of the existing sod. Generally, two specific conditions were responsible for poor establishment; either the terrain was too rough for proper drill function, or there was incomplete removal of the existing canopy of bermudagrass by mob grazing techniques before seeding. Of these, the latter could be better managed by close mowing before establishment as described by Hoveland et al. (1997); however, the rough terrain (rock outcroppings) on several of these experimental pastures prohibited this approach.

Forage Mass and Nutritive Value

Forage Mass

A critical management goal during the trial was to prevent overgrazing of the perennial cool-season forages. Bermudagrass is a C_4 forage that has a higher photosynthetic rate and efficiency at high radiation than C_3 forages (Nelson, 1995), but also is less tolerant of shading in mixed pastures than upright C_3 forages (Hoveland et al., 1997). Under the stressful climatic conditions of the southern Ozarks, avoiding overgrazing is critical for the persistence of E− and OG because it preserves the stem and tiller bases, which are the primary storage organs for carbohydrate reserves (Nelson, 1995). In addition, overgrazing allows more light to penetrate to the soil surface, thereby increasing the competitiveness of bermudagrass. Over the entire 4-yr trial, the mean forage mass was 3809 kg ha^{-1}; this was more than twice that reported by Hoveland et al. (1997) for pastures of E− overseeded into common bermudagrass and grazed with a rotational management strategy. We postulated that this conservative management approach was essential for persistence of both E− and OG, but especially for OG, which is known to be less tolerant of heat (Baker and Jung, 1968) and drought (Ball et al., 2002) than tall fescue.

Overall, forage mass was affected by the interaction of forage system × sampling date ($P = 0.001$); however, a closer inspection of these data indicated that forage mass changed over sampling dates in similar patterns for all forage systems. The interaction of forage system and sampling date was probably created by superior forage production for OG and E+ pastures relative to E− pastures during September 2002, and superior production from E+ and E− pastures compared to OG during June, July, and August 2003 (data not shown). This conclusion was supported by an additional statistical analysis (ANOVA) by year, in which no interaction of main effects was detected ($P \geq 0.727$) for either 2000 or 2001. Because the overall interaction of main effects was not likely related to divergent patterns of forage production for individual forage systems across sampling dates, the interaction will be ignored to permit a concise presentation of results; therefore, only main effect means of sampling date ($P < 0.0001$) are presented (Table 3) and discussed.

During 2000, excellent June rainfall (Table 2) resulted in a peak forage mass (5689 kg ha^{-1}; $P < 0.10$) in early July that declined ($P < 0.10$) during three of the five remaining monthly sampling intervals during that cal-

Table 2. Thirty-year normals (1971 to 2000) for precipitation and temperature, as well as cumulative monthly rainfalls and mean temperatures from 2000 through 2004 for the pasture site located near Batesville, AR.

	2000		2001		2002		2003		30-yr normal†	
Month	Precip.	Temp.	Precip.	Temp.	Precip.	Temp.	Precip.	Temp.	Precip.	Temp.
	mm	°C	mm	°C	mm	°C	mm	°C	mm	°C
Jan.	146	5.4	31	3.5	133	6.3	30	3.0	77	3.6
Feb.	67	10.3	166	7.5	52	6.0	111	4.6	84	6.7
Mar.	57	13.1	68	9.2	263	9.0	36	11.2	116	11.3
Apr.	68	16.1	27	19.2	50	18.2	59	17.6	112	16.3
May	154	22.0	71	21.4	91	19.8	270	20.6	122	20.4
June	108	24.3	65	24.8	108	25.0	142	22.7	86	24.9
July	24	27.0	126	28.8	63	27.4	144	26.6	80	27.7
Aug.	3	30.4	11	28.6	54	27.4	111	27.0	79	27.0
Sep.	52	24.5	98	22.3	83	24.6	96	20.8	94	23.1
Oct.	35	18.7	100	16.5	49	16.3	63	16.6	99	17.3
Nov.	161	8.9	146	13.3	48	9.9	153	11.7	135	10.7
Dec.	50	−0.7	247	7.2	195	6.6	114	5.0	102	5.3
Total	925	16.7	1156	16.9	1189	16.4	1329	15.6	1186	16.2

† Source: NOAA (2002).

Table 3. Forage mass over 41 sampling dates for mixed species pastures of orchardgrass (OG), endophyte-free tall fescue (E−), or endophyte-infected tall fescue (E+) with common bermudagrass. Pastures were grazed with either twice weekly (2W) or twice monthly (2M) rotation schedules near Batesville, AR (2000 to 2003).

	Year			
Sampling month	2000	2001	2002	2003
	kg ha^{-1}			
Jan.	2949	–†	–	3695
Feb.	2216	–	–	–
Mar.	2224	–	1967	–
Apr.	2971	1182	1977	3297
May	2215	2906	3962	3199
June	2899	3343	5928	4215
July	5689	2909	7290	5203
Aug.	4925	3886	5148	5632
Sept.	4829	3086	5707	4677
Oct.	3600	3413	4169	5505
Nov.	3644	4510	3725	4883
Dec.	2089	2438	4020	5245
LSD (0.10)‡	471.4			

† Pastures were not sampled during periods of winter hay feeding.
‡ Appropriate LSD (0.10) for comparing any two subplot (sampling date) means, regardless of column.

endar year. For 2001, forage mass peaked ($P < 0.10$) in November at 4510 kg ha^{-1} following slightly above normal precipitation during September and October; however, forage mass was likely limited during the spring and summer by below normal precipitation that occurred for all months from March through June. Among these, rainfall during April 2001 was only 24% of the 30-yr normal for that month (NOAA, 2002). During 2002, forage mass peaked in July at 7290 kg ha^{-1} following above average precipitation during June, and exceeded ($P < 0.10$) that observed on all other sampling dates by ≥1362 kg ha^{-1}; forage mass declined ($P < 0.10$) erratically thereafter, but remained at 3695 kg ha^{-1} as late as January 2003. Forage mass during 2003 was ≥4215 kg ha^{-1} from June through the conclusion of the trial; from May through December of that year, precipitation exceeded the 30-yr normal by 37%, and in all months except October.

In Vitro Dry Matter Disappearance

Concentrations of IVDMD varied with effects of sampling date ($P < 0.0001$), but not with any other main effect or interaction ($P \geq 0.105$). Averaged over all forage systems and sampling dates, the mean concentration of IVDMD was 586 g kg^{-1}. Concentrations of IVDMD reached a numerical maximum between 700 and 800 g kg^{-1} in April or May of 2000, 2001, and 2003 (Table 4), and then declined ($P < 0.10$) throughout the summer as cool-season plants aged and bermudagrass comprised a greater percentage of the forage mass. The trend was similar in 2002 except that the numerical maximum was only 685 g kg^{-1}.

Crude Protein

Concentrations of CP reached a numerical maximum ≥ 213 g kg^{-1} in March or April of each year (Table 5); in contrast, minimum concentrations of 93 and 99 g kg^{-1} were observed in September of 2000 and 2002, respectively. Overall, the mean concentration of CP was 157 g kg^{-1}, which is more than adequate to support the needs of either lactating or nonlactating beef cows (NRC, 1996), and likely reflects the aggressive annual N fertilization management (202 kg N ha^{-1}) utilized during the trial. Generally, concentrations of CP followed closely the trends described for IVDMD, exhibiting effects of only sampling date ($P < 0.0001$), but not forage system ($P = 0.933$) or the interaction of main effects ($P = 0.790$).

Table 4. Concentrations of in vitro dry matter disappearance (IVDMD) over 41 sampling dates for mixed species pastures of orchardgrass (OG), endophyte-free tall fescue (E−), or endophyte-infected tall fescue (E+) with common bermudagrass. Pastures were grazed with either twice weekly (2W) or twice monthly (2M) rotation schedules near Batesville, AR (2000 to 2003). Sampling date affected ($P < 0.0001$) concentrations of IVDMD but forage system ($P = 0.861$) and the interaction of forage system with sampling date did not ($P = 0.105$).

	Year			
Sampling month	2000	2001	2002	2003
	g kg^{-1}			
Jan.	599	–†	–	490
Feb.	508	–	–	–
Mar.	657	–	622	–
Apr.	779	682	685	772
May	611	738	684	666
June	572	614	549	582
July	607	594	553	619
Aug.	502	604	616	570
Sept.	523	597	516	518
Oct.	542	637	497	524
Nov.	534	620	525	517
Dec.	518	499	496	440
LSD (0.10)‡	29.3			

† Pastures were not sampled during periods of winter hay feeding.
‡ Appropriate LSD (0.10) for comparing any two subplot (sampling date) means, regardless of column.

Table 5. Concentrations of crude protein (CP) over 41 sampling dates for mixed species pastures of orchardgrass (OG), endophyte-free tall fescue (E−), or endophyte-infected tall fescue (E+) with common bermudagrass. Pastures were grazed with twice weekly (2W) or twice monthly (2M) rotation schedules near Batesville, AR (2000 to 2003). Sampling date affected ($P < 0.0001$) concentrations of CP, but forage system ($P = 0.933$) and the interaction of forage system with sampling date did not ($P = 0.790$).

	Year			
Sampling month	2000	2001	2002	2003
	g kg^{-1}			
Jan.	146	–†	–	125
Feb.	118	–	–	–
Mar.	213	–	200	–
Apr.	208	248	260	235
May	148	179	176	141
June	173	148	123	102
July	153	176	121	165
Aug.	118	182	122	133
Sept.	93	138	99	133
Oct.	150	179	155	146
Nov.	156	179	153	138
Dec.	152	167	132	127
LSD (0.10)‡	12.6			

† Pastures were not sampled during periods of winter hay feeding.
‡ Appropriate LSD (0.10) for comparing any two subplot (sampling date) means, regardless of column.

Species Composition and Basal Cover

Cool-Season Forages

Frequencies of OG, E−, and E+ exhibited an interaction of forage system and year ($P = 0.004$). Within all five forage systems, species frequencies varied ($P < 0.10$) across years; however, the overall patterns of change varied distinctly with forage system (Table 6). For OG2W, species frequency peaked numerically in 2001 at 52%, but this percentage did not differ ($P > 0.10$) from that observed for 2002. During 2003, there was a decline ($P < 0.10$) in OG frequency relative to that observed in 2001 and 2002; however, the percentage of OG for 2003 (39%) did not differ ($P > 0.10$) from that observed immediately before grazing was initiated in November 1999. For OG2M, frequencies peaked numerically at 42% during 2000, but did not differ ($P > 0.10$) from that exhibited in either 1999 or 2001. As observed for OG2W, a sharp decline ($P < 0.10$) in OG frequency occurred late in the trial; the observed frequency during both 2002 and 2003 was 24%, which represented a decline of 18 percentage units relative to the peak frequency observed during 2000. Generally, data for OG forage systems suggest that the shorter rest periods between grazing events for 2M was not adequate to maintain acceptable stands of OG more than four full years after establishment, but that acceptable OG stands may be maintained longer with the longer rest periods associated with the 2W rotation schedule (Table 1).

For E+2M, the frequency of E+ did not differ ($P > 0.10$) from November 1999 through 2002; the mean frequency over this time interval was 51%. The maximum frequency occurred in 2003 (61%), which differed ($P < 0.10$) from all other years. For both E−2W and E−2M, the frequency of tall fescue plants was numerically lowest in November 1999 (49 and 54%, respectively). Within these forage systems, frequencies increased ($P < 0.10$) to respective maxima of 71 and 72%; however, the maximum frequency for E−2M did not differ ($P > 0.10$) from frequencies observed on any other date after grazing was initiated. For E−2W, the frequency of E− varied between 61 and 71% after grazing was initiated, but only the frequency for 2001 was similar ($P > 0.10$) to the maximum. While these results are encouraging, they are somewhat in contrast with those of Hoveland et al. (1997). In that study, the frequency of E− increased over time in mixed pastures with bermudagrass when a rotational stocking strategy was used, but declined with continuous stocking management; however, the frequency of E− never exceeded 50% at any time during the trial, regardless of grazing management. A combination of factors likely can explain the excellent persistence of tall fescue in our E− pastures; these include: (i) aggressive fertilization with N timed to give E− a competitive advantage over bermudagrass; (ii) strict avoidance of overgrazing by maintaining a conservative forage mass (overall mean for all pastures = 3809 kg ha^{-1}) that limited the competitiveness of bermudagrass via shading; (iii) use of grazing management schemes that provided at least 15 d of rest between grazing events; and (iv) confining tractor and cow traffic during hay feeding.

Table 6. Frequencies of orchardgrass (OG), endophyte-free tall fescue (E−), or endophyte-infected tall fescue (E+) forages in mixed species pastures with common bermudagrass. Pastures were grazed with twice weekly (2W) or twice monthly (2M) rotation schedules near Batesville, AR (1999 to 2003). Pastures were evaluated before grazing was initiated (November 1999) and in June and November of each subsequent year. For 2000 through 2003, frequencies of the desired cool-season grass (OG, E−, or E+) and bermudagrass for each pasture represent the mean of evaluations made in June and November of each calendar year.

	Forage system				
Year	OG2W	OG2M	E+2M	E−2W	E−2M
	%				
Cool-season grass†					
1999	37c‡	36a	49b	49c	54b
2000	43bc	42a	54b	61b	65a
2001	52a	36a	52b	65ab	71a
2002	50ab	24b	50b	60bc	67a
2003	39c	24b	61a	71a	72a
SEM§	3.4	4.1	2.9	4.1	4.1
Bermudagrass					
1999	41ab	46bc	30b	36ab	32
2000	41ab	41c	28b	31ab	32
2001	35b	46bc	29b	27b	27
2002	36b	53ab	37a	37a	32
2003	47a	58a	30b	28b	27
SEM§	2.9	3.5	2.5	3.5	3.5

† No distinction has been made for the endophyte status of tall fescue plants in E+ or E− pastures. Each mean represents the frequency of plants in each pasture identified visually as tall fescue.

‡ Means within a column and forage type without common letters differ ($P < 0.10$).

§ Standard error of the forage system × year interaction mean.

Bermudagrass

For frequencies of bermudagrass (Table 6), there was a tendency ($P = 0.073$) for an interaction of forage system with year. Within OG2M pastures, bermudagrass frequencies ranged from 41 to 58%, and generally increased following 2000, averaging 52% from 2001 through 2003. Frequencies of bermudagrass in OG2W pastures increased sharply ($P < 0.10$) by 11 percentage units in 2003 from numerical minima (35 and 36%) observed in 2001 and 2002, respectively. Over the entire trial, the mean frequency of bermudagrass in OG2M was 9 percentage units less than observed during 5 yr for OG2W. For both OG systems, there was an inverse relationship between frequencies of OG and bermudagrass, and these data suggest that bermudagrass was opportunistic, readily filling in areas vacated by declining frequencies of OG (Table 6).

Within E+2M and E−2W pastures, frequencies of bermudagrass varied ($P < 0.10$) over dates, but these differences did not represent a clearly defined pattern, and were confined to relatively narrow ranges (28–37 and 27–37%, respectively; Table 6). For E−2M pastures, bermudagrass did not differ ($P > 0.10$) over years, averaging 30% for the entire trial. Generally, the relatively static responses of bermudagrass in all tall fescue pastures indicates that bermudagrass was effectively controlled by management practices that promoted shading of the soil surface.

Rogue Fescue Plants

Neither rotation frequency ($P = 0.522$) or the interaction of rotation frequency and year ($P = 0.160$) affected percentages of rogue tall fescue plants within OG pastures. However, frequencies of rogue fescue plants (Table 7) increased ($P < 0.10$) over years from a numerical minimum of 4% in November 1999 to a maximum of 10% during 2003, which was greater ($P < 0.10$) than observed for all other years except 2002 (8%; $P > 0.10$). Within E− pastures, no treatment effect was significant ($P \geq 0.209$), and the mean frequency from 1999 through 2001 was 2%. Generally, these findings are consistent with a recent report (Tracy and Renne, 2005) that found frequencies of E+ did not increase in pastures renovated with E− and other species over a 3-yr period in which they were grazed under rotational stocking.

These data suggest that a single application of glyphosate in the early spring provided good, but not complete, control of existing fescue plants. Previously, Defelice and Henning (1990) reported that spring applications of glyphosate at rates 1.68 and 2.52 kg a.i. ha^{-1} resulted in 88 and 93% visual control. However, it also was concluded that herbicide treatments were ineffective in killing old stands of E+ because still-active rhizomes from visually killed plants were a significant source of reinfection in new stands of E−. It is well known (Hill et al., 1991; Bouton et al., 1993; West et al., 1993; Malinowski and Belesky, 2000) that E+ plants possess physiological traits that should enable them to be more competitive in mixed stands than E−, thereby resulting in increased rates of infection over time. Previously, Shelby and Dalrymple (1993) reported reinfection rates of 4% per annum in Alabama pastures renovated with E−.

While there were rogue fescue plants that escaped herbicide kill scattered throughout most of the OG and E− pastures, their frequency was neither uniform across nor within individual pastures. Overall, frequencies of rogue fescue plants within individual OG and E− pastures ranged from 0 to 19%; however, greater frequencies were found most commonly in low-lying areas that tended to remain hydrated during periods of heat and drought stress. These observations were especially obvious for OG; three pastures exhibited a mean frequency across 5 yr of 2%, which agrees closely with that observed for E− pastures, and suggests that there was little reinfection by rogue E+ plants during the entire trial. The other two OG pastures contained low-lying areas (≈15% of the total pasture area), and frequencies within these pastures averaged about 13% over the entire trial, ranging from about 10% in 1999 to 17% in 2003. While this clearly indicates that rogue E+ plants spread within OG pastures, it should be noted that frequencies were calculated on the basis of the entire pasture area; high frequencies of rogue fescue plants were extremely localized within low-lying areas, and reestablishment of presumed E+ plants appeared to occur at a much faster rate within these areas than across the pasture generally.

Table 7. Frequencies of rogue tall fescue plants in common bermudagrass pastures overseeded with orchardgrass (OG) and endophyte-free (E−) tall fescue. Pastures were evaluated before grazing was initiated (November 1999) and in June and November of each subsequent year. For 2000 through 2003, frequencies of rogue tall fescue plants within each pasture represent the mean of evaluations made in June and November of each calendar.

Year	Seeded forage: OG	Seeded forage: E−†
	%	%
1999	4c‡	3
2000	5c	2
2001	6bc	2
2002	8ab	–
2003	10a	–
SEM§	1.0	0.5

† Within E− pastures, rogue tall fescue plants were identified based on crown size and orientation relative to the drill rows through 2001; after that time, they could not be distinguished from seeded plants.
‡ Means in a column without common letters differ ($P < 0.10$).
§ Standard error of the annual mean.

Other Species

A wide variety of other species were found throughout the study. Among these, the most common were little barley (*Hordeum pusillum* Nutt.), Kentucky bluegrass (*Poa pratensis* L.), southern crabgrass [*Digitaria ciliaris* (Retz.) Koel.], knotroot foxtail [*Setaria geniculata* (Lam.) Beauv.], johnsongrass [*Sorghum halepense* (L.) Pers.], dallisgrass (*Paspalum dilatatum* Poir.), chickweed [*Stellaria media* (L.) Vill.], henbit (*Lamium amplexicaule* L.), and annual ryegrass. Frequency of other species was affected only by year ($P = 0.001$). Estimates declined ($P < 0.10$) over years (Table 8), ranging from 18% in November 1999 down to 9% during 2003; however, the greatest decline (5 percentage units; $P < 0.10$) occurred during 2000, with only slow changes thereafter.

Basal Cover

Only the main effect of year affected ($P = 0.001$) estimates of basal cover. Estimates (Table 8) averaged 44% over the entire trial, ranging from 41 to 48%. Once grazing was initiated in January 2000, basal cover did not

Table 8. Frequencies of other species and percentages of total basal cover within mixed species pastures that contained common bermudagrass with either endophyte-free tall fescue (E−), orchardgrass (OG), or endophyte-infected tall fescue (E+). Pastures were evaluated before grazing was initiated (November 1999) and in June and November of each subsequent year. For 2000 through 2003, frequencies of other species and percentages of basal cover for each pasture represent the mean of evaluations made in June and November of each calendar year.

Year	Other species†	Basal cover
	%	%
1999	18a‡	45b
2000	13b	41c
2001	12b	42bc
2002	11bc	43bc
2003	9c	48a
SEM§	1.4	1.1

† Forage species other than the desired perennial cool-season grass (OG, E+, or E−) and bermudagrass.
‡ Means in a column without common letters differ ($P < 0.10$).
§ Standard error of the annual mean.

differ through 2002 (overall mean = 42%; $P > 0.10$); however, a 5 percentage unit increase ($P < 0.10$) occurred for 2003, but this was likely related to the favorable rainfall patterns observed throughout that summer (Table 2). Generally, the common bermudagrass base within these pastures was quite vigorous, and exhibited dense sod characteristics. Although estimates of basal cover varied ($P < 0.10$) over years, there was no suggestion it was either improved or reduced by any of the forage systems evaluated.

Total Ergot Alkaloids and Percent Endophyte Infection

Total Ergot Alkaloids

Within OG and E+ pastures, concentrations of ergot alkaloids were affected by the interaction of main effects ($P = 0.001$). Within OG2M and E+2M pastures, concentrations of ergot alkaloids were greater ($P < 0.10$) in both 2002 and 2003 than in either 2000 or 2001 (Table 9). For both forage systems, there was an approximate threefold increase in ergot alkaloids during the last 2 yr of the trial, relative to the first 2 yr. In contrast, concentrations of ergot alkaloids within OG2W pastures varied ($P < 0.10$) erratically over years, which likely contributed to the interaction.

During 2000 and 2001, most E− pastures had very low estimates of frequency for rogue fescue plants (Table 7), and in some pastures there were none identified. This also was true during sampling for total ergot alkaloids, and created a procedural problem; therefore, when no rogue plants could be identified, all fescue plants were sampled at random as an independent corroboration of low endophyte infection. This alternative sampling procedure was used for one E− pasture in 2000 and for two pastures in 2001. Concentrations of total ergot alkaloids in all three cases were not detectable. For the remainder of the E− pastures, there were no effects ($P \geq 0.527$) of rotation frequency during this time period, and the overall mean concentration of total ergot alkaloids in rogue fescue plants was 472 ng g^{-1} (SEM = 50.9 ng g^{-1}).

During 2002 and 2003, all fescue plants in E− pastures were sampled because rogue plants could no longer be identified visually. There was a tendency for an effect of year ($P = 0.055$) during this time period, but a single extreme value may have contributed heavily to this observation. Over the final 2 yr of the trial, the mean concentration of total ergot alkaloids in E− pastures was 523 ng g^{-1} (SEM = 68.8 ng g^{-1}), which was approximately 25% of that observed for E+ and OG pastures. For purposes of interpretation, it should be noted that concentrations of total ergot alkaloids were determined for tall fescue plants only; dilution effects by other forage species are not reflected in these results.

Since glyphosate was applied only once before establishment of E− and OG pastures, complete control of preexisting and presumably E+ plants was not expected (Defelice and Henning, 1990), and did not occur (Table 7). In part, reinfection of recently renovated E− pastures with E+ may occur because mature, rhizomatous plants escape herbicide kill (Defelice and Henning, 1990), and because survival rates for E+ and E− plants may differ (Shelby and Dalrymple, 1993). Clearly, reinfection occurred by these or other mechanisms because measurable concentrations of ergot alkaloids were found in E− pastures in both 2002 and 2003. However, we are not aware of other grazing studies with other treatments (OG) in which rogue fescue plants could be counted visually. Theoretically, reinfection should occur by identical mechanisms within both E− and OG pastures. Considering the widely held view throughout the southern Ozarks that OG is less persistent than E−, it is reasonable to expect that E+ plants would increase even more rapidly in OG than in E− pastures. These data suggest that reinfection of OG occurred at a relatively slow rate during the trial, reaching a maximum frequency of 10% during the final year of the trial, but averaging only 6% over the previous 4 yr (Table 7). Furthermore, livestock performance did not suggest that there were large increases in pasture toxicity. Mean annual weaning weights for calves raised across all pastures ranged only slightly from 246 to 250 kg during the trial, and effects of year and forage system × year were not significant (Coblentz et al., 2006). Depressions in livestock performance (Paterson et al., 1995) and significant effects of year and/or interactions with year would be expected with increases in pasture toxicity.

Table 9. Concentrations of total ergot alkaloids in tall fescue plants sampled from orchardgrass pastures rotated twice weekly (OG2W) or twice monthly (OG2M), and endophyte-infected tall fescue pastures rotated twice monthly (E+2M).

	Forage system†		
Evaluation date	OG2W	OG2M	E+2M
	ng g^{-1}		
June 2000	1125b‡	417b	565c
June 2001	1306b	894b	1009b
June 2002	859b	2048a	2279a
June 2003	1849a	1921a	2023a
SEM§	189.0	231.5	163.7

† Alternate sampling procedures were used to assess concentrations of ergot alkaloids in endopyte-free (E−) tall fescue pastures. Data are summarized in the text.
‡ Means in a column without common superscripts differ ($P < 0.10$).
§ Standard error of the forage system × sampling date interaction mean.

IMPLICATIONS

Generally, the rotational stocking strategies used in these studies coupled with the strict avoidance of overgrazing were effective at maintaining populations of E− and OG, and at limiting the competitiveness of the base sod of bermudagrass. Conditions regulating reinfection of renovated pastures remain incompletely defined, and the threshold level of toxicity and/or dilution required to affect livestock performance either negatively or positively remains equally unclear. These results suggest that pasture toxicity can be reduced substantially for at least 5 yr in the southern Ozarks with a single application of glyphosate followed by establishment of nontoxic perennial cool-season forages.

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